MICROSCOPY.[1]

Methods of Preparing Molluscan Ova.[2]—The results of my work for the first three months were not promising and it was not until I had hit upon my present methods of preparing surface views of the entire ova that any detailed study of the cleavage could be made. Since I owe most of my results to this method and since I am convinced that it may be profitably employed in the preparation of the surface views of many different objects I believe it merits a detailed description.

The ova were fixed in many different fluids—Kleinenberg's Picro-sulphuric, Picric acid in sea water, Merkel's, Perenyi's, Flemming, stronger and weaker, Auerbach's, Corrosive sublimate, Chromo-formic, Chromo-acetic and absolute alcohol—but none of these methods for a moment compare with the first named, i. e. Kleinenberg's stronger picro-sulphuric. The ova were left in this for a length of time varying from fifteen minutes to one hour and were then gradually transferred to 70% alcohol. They were left in this until all traces of picric acid had been washed out and were finally preserved in 95% alcohol. During the first year of the work many of the preparations were ruined by becoming very dark, owing I think to the extraction of tannin from the corks. This trouble was afterward avoided by using rubber corks, or better still by coating ordinary corks with a thin layer of paraffin.

As a result of many experiments with almost every one of the common staining fluids, I found that the best method of preparing surface views of the whole egg or embryo was the following:—(1) Transfer the object gradually from alcohol to water. (2) Stain from five to ten minutes in a solution of Delafield's (Grenacher's) Hæmatoxylin diluted about six times with distilled water and rendered *slightly* acid by a trace of HCl. (3) Dehydrate and clear in oil of cedar or cloves. (4) Mount in Balsam supporting the cover glass so as to prevent crushing. By occasionally softening the balsam with a drop or two of xylol and slightly moving the cover glass the objects can be rolled into any position desired.

By this method wonderfully beautiful surface preparations were obtained showing with remarkable clearness not only the nuclei and cell boundaries but also the caryokinetic figures and in many cases the archoplasmic spheres and centrosomes. One very considerable advan-

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([2]Extracted from a paper to be published later on the development.)

tage of this method is that the preparations were permanent—in fact they become better with age instead of degenerating. All the preparations from which the figures were drawn are still in existence and can be consulted at any time.

I have employed this method with almost as good results in the preparation of surface views of the embryo chick and English sparrow and also with considerable success on other molluscan eggs and embryos as well as those of annelids and echinoderms.

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